67

## **CLAIMS**

A method for the determination of water quality, which method comprises:
 contacting a vessel comprising a population of bacteria with a water sample
to be tested, said vessel comprising a semi-permeable material which allows the
water sample to pass therethrough and contact said bacteria; and

determining the growth rate of the bacteria and proportion of respiring bacteria in the vessel,

thereby to determine the water quality of the water sample.

- 2. A method according to claim 1, wherein a test sample is withdrawn from the vessel and the growth rate of the bacteria and proportion of respiring bacteria in the test sample is determined.
- 3. A method according to claim 1 or 2, wherein two or more vessels, each comprising a population of bacteria, are simultaneously contacted with the water sample to be tested.
- 4. A method according to any one of the preceding claims, wherein the semipermeable material is dialysis tubing.
- 5. A method according to any one of the preceding claims, wherein the semipermeable material has a molecular weight cut-off of from about 100Da to about 300kDa.
- 6. A method according to claim 5, wherein the semi-permeable material has a molecular weight cut-off of about 60kDa.
- 7. A method according to claim 5, wherein the semi-permeable material has a molecular weight cut-off of about 25kDa.

68

- 8. A method according to claim any one of the preceding claims, wherein the population of bacteria comprises one or more species of marine or freshwater bacteria or a mixture of any thereof.
- 9. A method according to any one of the preceding claims, wherein the population of bacteria comprises genetically modified bacteria.
- 10. A method according to claim 8 or 9, wherein the population of bacteria comprises one or more *Pseudomonas* and/or *Vibrio* species.
- 11. A method according to any one of the preceding claims, wherein the population of bacteria is in the exponential growth phase when the vessel is contacted with the water sample to be tested.
- 12. A method according to any one of claims 2 to 11, wherein more than one test sample is withdrawn from the or each vessel, each test sample being withdrawn at a different predetermined time point.
- 13. A method according to claim 12, wherein a first sample is withdrawn within ½h of the contacting step, a second sample is withdrawn at about 6h after the contacting step and a third sample is withdrawn at about 24h after the contacting step.
- 14. A method according to claim 12 or 13, wherein a first sample is withdrawn within ½h of the contacting step and a further sample is withdrawn at each hour after the contacting step up to 24h after the contacting step.
- 15. A method according to any one of the preceding claims, wherein the growth rate of the bacteria in the vessel or test sample is determined by determining the optical density and/or the turbidity of the bacterial population in the vessel or test sample.

69

- 16. A method according to claim 15, wherein the optical density is determined at a wavelength of about 540nm.
- 17. A method according to any one of the preceding claims, wherein the proportion of respiring bacteria in the vessel or test sample is determined by contacting the bacterial population in the vessel or test sample with a redox dye and determining the level of fluorescence of the resulting bacterial population in the vessel or test sample.
- 18. A method according to claim 17, wherein the level of fluorescence of the resulting bacterial population in the vessel or test sample is determined by exciting the bacterial population in the vessel or test sample using light of a predetermined intensity at a first predetermined wavelength and measuring the intensity of fluorescence at a second predetermined wavelength.
- 19. A method according to claim 17 or 18, wherein the redox dye is 5-cyano-2,3-ditolyl tetrazolium chloride.
- 20. A method according to any one of the preceding claims, wherein a decrease in bacterial growth rate and/or in the proportion of respiring bacteria in the vessel of test sample as compared to the corresponding bacterial growth rate and/or proportion of respiring bacterial in a control water sample is indicative of a decrease in water quality.
- 21. A method according to any one of the preceding claims which is adapted to be carried out in the field.
- 22. A detection device for use in a method according to any one of the preceding claims which device is arranged to determine the optical density and/or turbidity of a bacterial culture and also to determine the level of fluorescence of a bacterial culture contacted with a redox dye.

PCT/GB2004/004189

- 23. A detection device according to claim 22, said device comprising means for determining the optical density of a bacterial culture at a wavelength of about 540nm.
- 24. A detection device according to claim 22 or 23, comprising a photodiode detector for determining the optical density of a bacterial culture.
- 25. A detection device according to any one of claims 22 to 24, said device comprising means for determining the level of fluorescence of a bacterial culture contacted with a redox dye by exciting the culture using light of a predetermined intensity at a first predetermined wavelength and measuring the intensity of fluorescence at a second predetermined wavelength.
- 26. A detection device according to claim 25, wherein said means for determining the level of fluorescence of a bacterial culture contacted with a redox dye comprises a light source for emitting said light of said first predetermined wavelength and a detector for detecting light of said second predetermined wavelength.
- 27. A detection device according to claim 25 or 26, wherein the first predetermined wavelength is about 488nm and the second predetermined wavelength is about 630nm.
- 28. A detection device according to any one of claims 22 to 27, which is a hand-held device.
- 29. A kit of components for use in carrying out a method according to any one of claims 1 to 21, which kit of components comprises:

one or more vessels comprising a semi-permeable material which allows a water sample to pass therethrough;

a non-watertight carrying structure to carry the one or more vessels; and

71

a member for securing the position of the one or more vessels within the carrying structure and/or for providing flotation of the carrying structure.

- 30. A kit of components according to claim 29, wherein said member is a buoyant member for providing flotation of the whole kit of components.
- 31. A kit of components according to claim 29 or 30, wherein the one or more vessels are each dialysis tubes.
- 32. A kit of components according to any one of claims 29 to 31 which further comprises a detection device according to any one of claims 22 to 28.
- 33. A kit of components according to any one of claims 29 to 32 which comprises means for transporting together the other components of the kit.
- 34. A kit of components according to any one of claims 29 to 33, wherein said non-watertight carrying structure is a basket.
- 35. A deployment device suitable for carrying one or more vessels that each comprise a semi-permeable material which allows a water sample to pass into said vessels, said deployment device comprising a non-watertight housing and means for securing the one or more vessels within the housing and/or means for providing flotation of the deployment device.
- 36. A sampling device for use in a method according to any one of claims 1 to 21, which sampling device comprises:
- (i) a sampling chamber which is adapted to accept one or more vessels comprising a semi-permeable material which allows a water sample to pass therethrough; and
- (ii) a closure member selectively switchable between two or more positions, wherein:

72

in one of the said positions the closure member seals the sampling chamber to prevent ingress of water into the sampling chamber when the device is submerged in water; and

in a second of the said positions the closure member is open to allow ingress of water into the sampling chamber when the device is submerged in water.

- 37. A sampling device according to claim 36, which comprises two or more sampling chambers, each with a closure member, wherein the sampling chambers are arranged so that each sampling chamber is positioned at a different depth when the sampling device is submerged in water.
- 38. A sampling device according to claim 36 or 37, wherein said sampling chambers are connected together by a cable so that said sampling device may be deployed by lowering said cable into water to position said two or more sampling chambers at respectively different depths.
- 39. A sampling device according to any one of claims 36 to 38, wherein said closure member is activated by electro-mechanical means.
- 40. A sampling device according to claim 39 when appendent on claim 38, wherein said cable is arranged to carry actuation signals to open or close said closure member.
- 41. A sampling device according to claim 39 or 40, wherein said closure member comprises a plate having one or more water ingress holes therethrough and one or more index holes therethrough, said plate being moveable so as to selectively open the sampling chamber in the second position by presenting a water ingress hole over said sampling chamber.
- 42. A sampling device according to claim 41, wherein said plate is spring-loaded and is indexed by a solenoid actuator cooperating with said one or more index holes.

73

43. A sampling device according to claim 41 or 42, further comprising a microswitch for providing feedback as to the open or closed state of said plate.